

Sperm Diffusion Models and *In Situ* Confirmation of Long-Distance Fertilization in the Free-Spawning Asteroid *Acanthaster planci*

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Abstract. This study was undertaken to compare fertilization rates of the sea star *Acanthaster planci* that were predicted using sperm diffusion models with those that were determined under natural conditions in the field. During experimentally induced spawnings, measured fertilization rates for broadcast eggs were high. More than 70% of the eggs were fertilized at distances as great as 8 m downstream from a single spawning male starfish, and more than 20% were fertilized at separations of more than 60 m. Fertilization was still measurable, at 5.8%, 100 m downstream. Lateral diffusion of sperm away from the axis of flow produced fertilization rates of 13.8% at 8 m normal to the flow and 32 m downstream. The large volumes of sperm released by male *A. planci* are the primary cause of high rates of fertilization for eggs derived from widely spaced individuals. Models of sperm diffusion using high sperm release rates such as those found in this starfish accurately confirmed the fertilization rates measured *in situ* for two populations of *A. planci* with widely differing rates of sperm release.

We observed some changes in starfish density and degree of aggregation in the study population for spawning periods during two spawning seasons, though these were not striking. High levels of aggregation may not be necessary for fertilization success in this starfish, due to the potential for long-distance fertilization and the probability that, for any spawning starfish, the total number of zygotes formed will be greater at some distance from the point of spawning. Although fertilization rates in areas distant from

the sperm source were relatively low, the total area for potential gamete encounters is much greater and may make a large contribution to net fertilization. We predict that other behaviors, such as migration to shallow water, commonly associated with spawning in *A. planci* and other marine invertebrates will have measurable impacts on fertilization success.

The potential for high levels of fertilization in *A. planci* was realized during natural spawnings. Fertilization rates as high as 99% were recorded when levels of spawning synchrony were high.

Introduction

Do free-spawning marine invertebrates ever achieve high rates of fertilization? Attempts to answer this question have increased in frequency and taken a variety of forms, most of which use echinoderms as conceptual and empirical models. The question arises for two main reasons. First, many—perhaps even most—observations of natural spawning have involved isolated or lone individuals (usually males) or only a small proportion of the population present. This is true for echinoids (*Diadema*: Randall *et al.*, 1964; Levitan, 1988; *Strongylocentrotus*: Pennington, 1985, Pearse *et al.*, 1988), holothuroids (Mosher, 1982; McEuen, 1988; Pearse *et al.*, 1988), and asteroids (Minchin, 1987; Pearse *et al.*, 1988; Babcock and Mundy, 1992; Gladstone, 1992). Similar behaviors have also been noted for other groups such as tridacnid clams (Braley, 1984) and sponges (Reiswig, 1970). Second, experimental studies of *in situ* fertilization rates for free-spawning species show that fertilization can be very low or even fail completely unless animals spawn with a high degree of synchrony and are in extremely close proximity (<1 m) during spawning (Pennington, 1985; Levitan, 1991; Levitan *et*

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al., 1992). Numerical modeling studies of sperm diffusion support these empirical results by predicting that concentrations of sperm will be too low to effect high levels of fertilization except when animals are close together (Denny and Shibata, 1989).

Nevertheless, the continued existence of thousands of such species in the seas today suggests that high levels of fertilization can, and do, occur in free-spawning marine invertebrates. It seems unlikely that so much effort would be devoted to gamete production instead of other functions if all the gametes were to be wasted. The strongest evidence that high rates of fertilization can be commonplace is indirect, coming from the evolution of a wide variety of mechanisms to prevent polyspermy (e.g., echinoids: Scheul, 1984). Although most observations have been of asynchronous or isolated spawnings, virtually all of the taxa listed above have been observed in the act of large-scale synchronous spawnings (Hoppe and Reichert, 1987; Reiswig, 1970; Pennington, 1985; Babcock and Mundy, 1992; McEuen, 1988; Minchin, 1987). Such large-scale synchronous spawnings are likely to be much more important to the reproductive output of the population than indicated by their low frequency.

For free-spawning marine organisms to attain high levels of fertilization, the numbers or concentration of viable sperm adjacent to eggs must be above a certain level. Two types of adult behavior, synchronization of spawning and aggregation of reproductive adults, can increase the concentration of sperm in space and time. A third strategy is to increase sperm output. Levitan (1991) postulated that, in terms of zygote production, "small organisms living at high population density may be just as fecund as large organisms living at low population density." That study was unable to demonstrate any effect of adult size on fertilization rate, though adult density was shown to have a major influence, with success ranging from 7.3% (with 1 male per square meter), to 45% (with 16 males per square meter).

Our study of *in situ* fertilization rates concentrated on the crown-of-thorns starfish, *Acanthaster planci*. This large (25–40 cm diameter), highly fecund asteroid is characterized by population outbreaks that have impacted large areas of coral reef throughout the Indo-Pacific region over the last 30 years (Moran, 1986). Several hypotheses have been formulated to explain these outbreaks. One set of theories proposes that they are a natural result of enhanced reproductive success of starfish in aggregations (Dana, 1970; Vine, 1970; Moore, 1978). Such aggregations can form naturally as a result of feeding behavior (Ormond et al., 1973) or environmental disturbances such as cyclones (Dana, 1970). Recent measurements of fertilization rates for *A. planci* suggest that the reproductive biology of *Acanthaster* populations may have unusual aspects that contribute to periodic huge increases in abundance. Fer-

tilization rates greater than 20% have been reported for eggs released 64 m downstream from a single spawning male *Acanthaster* (Babcock and Mundy, 1992). This level of fertilization is approximately equivalent to that found for spawning sea urchins at distances of less than 1 m (Pennington, 1985; Levitan, 1991). The scale at which *A. planci* is able to maintain reproductive contact is two orders of magnitude greater than that reported for urchins or any other marine invertebrates (e.g., hydroids: Yund, 1990; ascidians: Grosberg, 1991). The source of this difference could lie either in the greater size of *Acanthaster* and the large mass of gametes it releases (Babcock and Mundy, 1992) or in the physiology of the gametes.

We observed individuals in a population of starfish over two spawning seasons in an effort to determine whether, over the course of the spawning season, there were any differences in vertical distribution, degree of aggregation, or other factors that might enhance fertilization rates. Natural spawning events gave us an opportunity to measure fertilization rates *in situ* and assess the importance of spawning synchrony to fertilization success.

Combining empirical and modeling approaches, we attempted to determine whether number of gametes alone could adequately explain the ability of *Acanthaster* to fertilize at great distances. We also observed a natural population to assess how the results of modeling studies could be related to natural spawning behavior. In a series of field trials, we examined the effects of population density on fertilization success, and compared these directly with the results of modeling studies.

Materials and Methods

Behavioral observations

The population of starfish at Davies Reef (Lat. 18°50' S, Long. 147°39' E) in the central Great Barrier Reef was regularly sampled and observed visually over two spawning seasons (December–January) of 1990–91 and 1991–92. The observation site was in the lagoon of the reef, adjacent to the site of the fertilization experiments. Depths varied between 1 and 10 m, and many coral heads and patch reefs with abundant live coral were present. A belt transect 200 m long, 10 m wide and divided into 10 quadrats of equal size (20 m × 10 m) was established in the area. The number and behavior (spawning/not spawning) of starfish in each quadrat on the transect were recorded on 60 occasions (20 at night). A total of 214 dives (55 at night) were made on this population in the course of collecting these data and while making general observations or conducting fertilization experiments. All natural spawnings of *Acanthaster* were noted and, when females were observed spawning, eggs were collected to enable us to measure natural fertilization rates.

Induced spawning experiments

Rather than manipulating the density of populations (Levitan *et al.*, 1991), we varied the distance between spawning pairs of male and female starfish *in situ*, both along the direction of the current and normal to it. The low density of the starfish in the field and the large scale over which the experiment had to be conducted were logistic reasons for using pairs of animals rather than arrays of spawning animals. Currents prevailing during the experiments were of a sufficient velocity ($0.07\text{--}0.25\text{ m}\cdot\text{s}^{-1}$) that the major effects were likely to be observed downstream (Babcock and Mundy, 1992). The sex of animals to be used in the trials was determined in advance with a syringe and 14-gauge needle. Each animal was injected with $20\text{--}30\text{ ml } 10^{-4}\text{ M}$ 1-methyl adenine to induce spawning. Because they reacted more slowly, females were treated approximately 10–15 min before males. Most of the fertilization trials took place at Davies Reef in 1991–92; however an additional set of trials was run in June 1991 at Sesoko Island, Okinawa, and a few trials using increased numbers of spawning male starfish were also conducted at Davies Reef late in the 1991–92 spawning season.

Immediately after the starfish were injected with methyl adenine, current direction was determined by release of fluorescein dye. The male starfish was placed at the point of dye release and the female was moved a predetermined distance directly downstream. Divers monitored the male starfish, and released extra dye when sperm release was fully under way. Sampling of eggs commenced when the dye cloud reached the female, or as soon thereafter as the female began releasing eggs. The spawning female was incrementally moved upstream from the most distant sampling point, and eggs were sampled at progressively smaller distances downstream from the spawning male. Sampling points in the trials were 100 m, 64 m, 32 m, 16 m, 8 m, 4 m, 2 m, 0 m (adjacent downstream), as well as -4 m and -8 m (upstream). The exact sampling routine in each trial varied because it was not possible to sample every position each time. This sampling strategy allowed us to measure the effect of sperm transport and diffusion in a direct downstream direction. Starfish were also deployed on a grid of coordinates at various distances perpendicular to the flow to allow measurement of the effects of lateral sperm diffusion. At distances of 32, 16, 8, 4, 0, and -4 m , an additional series of samples was taken from females at varying distances normal to the flow (16, 8, 4, 2, and 0 m). Samples at Sesoko Island were taken only along the direct downstream vector, up to a distance of 32 m. In all cases, sampling was begun at the most distant point downstream and proceeded upstream past the spawning male animal, the last point acting as a sperm-free control. Immediately before and after each

spawning trial, current speed was measured using a tape, neutrally buoyant markers, and a stopwatch. The trials were planned so that the current would flow in a consistent direction for the duration of the trial.

Samples of eggs were collected using a submersible plankton pump equipped with $64\text{-}\mu\text{m}$ -mesh filter cartridges (Babcock and Mundy, 1992) that allowed the collection of 12 individually metered samples (1–21) during each trial. Once collected, these samples were completely isolated from water outside by means of one-way valves. Trials of this device demonstrated that neither the volume of water pumped through the filter cartridges (48 trials ranging from 0.5 to 3 l, $R^2 = 0.02$) nor the period of time eggs remained in contact with the water sample (2 min to about 1 h, $p = 0.95$), affected the eventual level of fertilization (Mundy *et al.*, unpublished data). Two samples were taken at each sampling point and preserved at the termination of the experiment. Fertilization of a subsample of the eggs was then scored in the laboratory.

Individual *A. planci* from both Davies Reef and Sesoko Island were sampled to determine the mean size and mean gonad weight of the two populations. To estimate the average mass of gametes spawned by Davies Reef starfish, we used mean gonad weights for the population immediately before and after natural spawning events observed on Davies Reef in 1990 and 1991.

Sperm diffusion modeling

The shedding of sperm into a flow was modeled as a plume diffusion problem in which the concentration of particles downstream from a point source was described under a set of simplifying assumptions (Csanady, 1973; Denny and Shibata, 1989). In this model the water column is assumed to be moving at a mean velocity $\bar{U}$ parallel to the x -axis within a turbulent benthic boundary layer. A point source shedding sperm at a constant rate Q_s located at the x, y origin, at a distance h , above the substratum gives a function for the concentration of *A. planci* sperm S ,

$$S(x, y, z) = \frac{Q_s}{2\pi\bar{U}\sigma_y\sigma_z} \exp \frac{-(y^2)}{2\sigma_y^2} \times \left\{ \exp \frac{-(z+h)}{2\sigma_z^2} + \exp \frac{-(z-h)}{2\sigma_z^2} \right\} \quad \text{Eq. 1.}$$

where the spatial standard deviations for diffusion are modeled by the functions

$$\sigma_y = \alpha_y \left(\frac{h^*}{\bar{U}} \right) x^\beta \quad \text{Eq. 2.}$$

$$\sigma_z = \alpha_z \left(\frac{h^*}{\bar{U}} \right) x^\beta \quad \text{Eq. 3.}$$

The values for the empirically determined diffusion parameters α_y , α_z describe the shape of the plume, and β describes the rate of growth of the eddies acting to disperse the plume as it spreads out. Without separate data from which to determine the α_z , α_y and β parameters, it was necessary to estimate their values by fitting the model to the fertilization success data. Ranges for suitable values of the plume shape parameters were suggested by Denny (1988). He suggested ranges of 0.5 to 3 for α_z , 1 to 3 for the ratio α_y/α_z , and 0.9 to 1.33 for β . The shear velocity u^* is a function of the Reynold's shear stress and is taken to be a measure of the turbulence intensity. It is used to describe the mean distance a particle is moved by the turbulent eddies over a small interval of time. The calculation of real values for the shear velocity is beyond the scope of this simple model, and we have adopted the values cited by Denny (1988) where the shear velocity is a constant fraction (10%) of the mean velocity.

At shallow depths the upper surface boundary is likely to play a significant role by restricting the dispersion of gametes. The original model (Csanady, 1973) was designed to work for atmospheric dispersion, recognising only a lower boundary to the flow. Given the depth of water at the field site (7 m) and the scale at which the effects of sperm diffusion were observed (100 m), it was necessary to include a term describing surface reflection. By adding a mirror source located above the sea-surface boundary, we reflect any sperm that diffuses as far as the surface back into the flow. In water of depth D , the resultant sperm concentration is

$$S = \frac{Q_s}{2\pi \bar{U} \sigma_y \sigma_z} \exp \frac{-(y^2)}{2\sigma_y^2} \left\{ \exp \frac{-(z+h)^2}{2\sigma_z^2} + \exp \frac{-(z-h)^2}{2\sigma_z^2} + \exp \frac{-[(2D-z-h)]^2}{2\sigma_z^2} \right\} \quad \text{Eq. 4.}$$

By calculating the probability of sperm colliding with ova (Vogel *et al.*, 1982; Denny and Shibata, 1989), the percent fertilization success at a point is determined from the sperm concentrations.

$$F(x, y, z) = 1 - \exp(-\phi t u^* S) \quad \text{Eq. 5.}$$

The shear velocity is used as a measure of the distance a sperm travels in unit time. Time t is the time spent observing collisions at a point. The area ϕ is $\sim 3\%$ of the cross-sectional area (*cf.* Levitan *et al.*, 1992) of an unfertilized *A. planci* egg 0.2 mm in diameter or approximately $9.42 \times 10^{-10} \text{ m}^2$. Because shear velocity is assumed to be a constant fraction of the mean velocity, all the velocity terms in the calculation of fertility success reduce to a constant. Although the calculated sperm concentrations vary with flow velocity, this variation is canceled by the u^* term in the calculation of fertilization success. This shortcoming could be remedied through models that bet-

ter relate shear velocity to flow velocity and by empirically relating fertilization success to sperm concentration.

Denny and Shibata (1989) followed a cohort of sperm and ova as it advected downstream. By iterating simultaneous differential equations for the concentrations of sperm and ova and calculating the predicted fertilization rate in a closed vessel (Vogel *et al.*, 1982), they were able to estimate fertilization rates and compare the results of their model with those of Pennington (1985) for *Strongylocentrotus*. Fertilization success for *A. planci* was measured by sampling eggs over a period of about 30 s (the time taken to sample 1–2 l water). In terms of our model, concentration and fertilization are calculated at discrete positions, with each point experiencing relatively constant sperm concentrations over the time taken to sample the eggs. We have not modeled the dispersion of ova, since sampling was in the free flow immediately downstream from spawning females. It is assumed that the ratio of sperm to ova is sufficient to ensure that the concentration of eggs has a negligible effect on the results. Because most fertilization is likely to take place in open water over the female starfish, very soon after the eggs and sperm mix (90% in the first 20 s; Denny and Shibata, 1988), we used the equation appropriate for estimating sperm-egg collisions in a turbulent flow, rather than that for an enclosed vessel (*i.e.*, Denny and Shibata 1988, p. 880).

Data collected from our *A. planci* population at Davies Reef before and after natural spawnings during the 1990–91 and 1991–92 spawning season (Babcock and Mundy, 1992; and unpublished data) gave an average gamete release for males of 60–106 g, shed over about 45 min. These values are about one-third of the prespawning gonad mass. We estimated the amount of gonad spawned by male starfish at Sesoko Island as 14 g, based on shedding of one-third of the gonad and measurements of total gonad weight. Male starfish from Davies Reef were 2.8 times larger than those at Sesoko Island (mean whole wet-weights: Davies Reef 2220 g, Sesoko Island 780 g). Assuming that the concentration of undiluted *A. planci* sperm is equal to that of urchin sperm ($2 \times 10^{16} \text{ sperm} \cdot \text{m}^{-3}$; Tyler *et al.*, 1956) and that the sperm are essentially neutrally buoyant, the release rate, Q_s , of sperm is $4.44\text{--}7.84 \times 10^8 \text{ sperm} \cdot \text{s}^{-1}$. We used a mean sperm release rate per animal of $6.14 \times 10^8 \text{ sperm} \cdot \text{s}^{-1}$ for Davies Reef starfish, and $1.5 \times 10^8 \text{ sperm} \cdot \text{s}^{-1}$ for Sesoko Island starfish.

The model assumes a constant flow velocity throughout the water column. Flow velocities recorded at the field site during experiments ranged from 0.07 to $0.25 \text{ m} \cdot \text{s}^{-1}$ with a mean velocity $\bar{U}$ of $0.12 \text{ m} \cdot \text{s}^{-1}$. The high degree of mixing in a turbulent boundary layer allows us to make this assumption except near the substratum, where the assumption fails because the flow velocity must reduce to zero. It is assumed that sampling takes place far enough

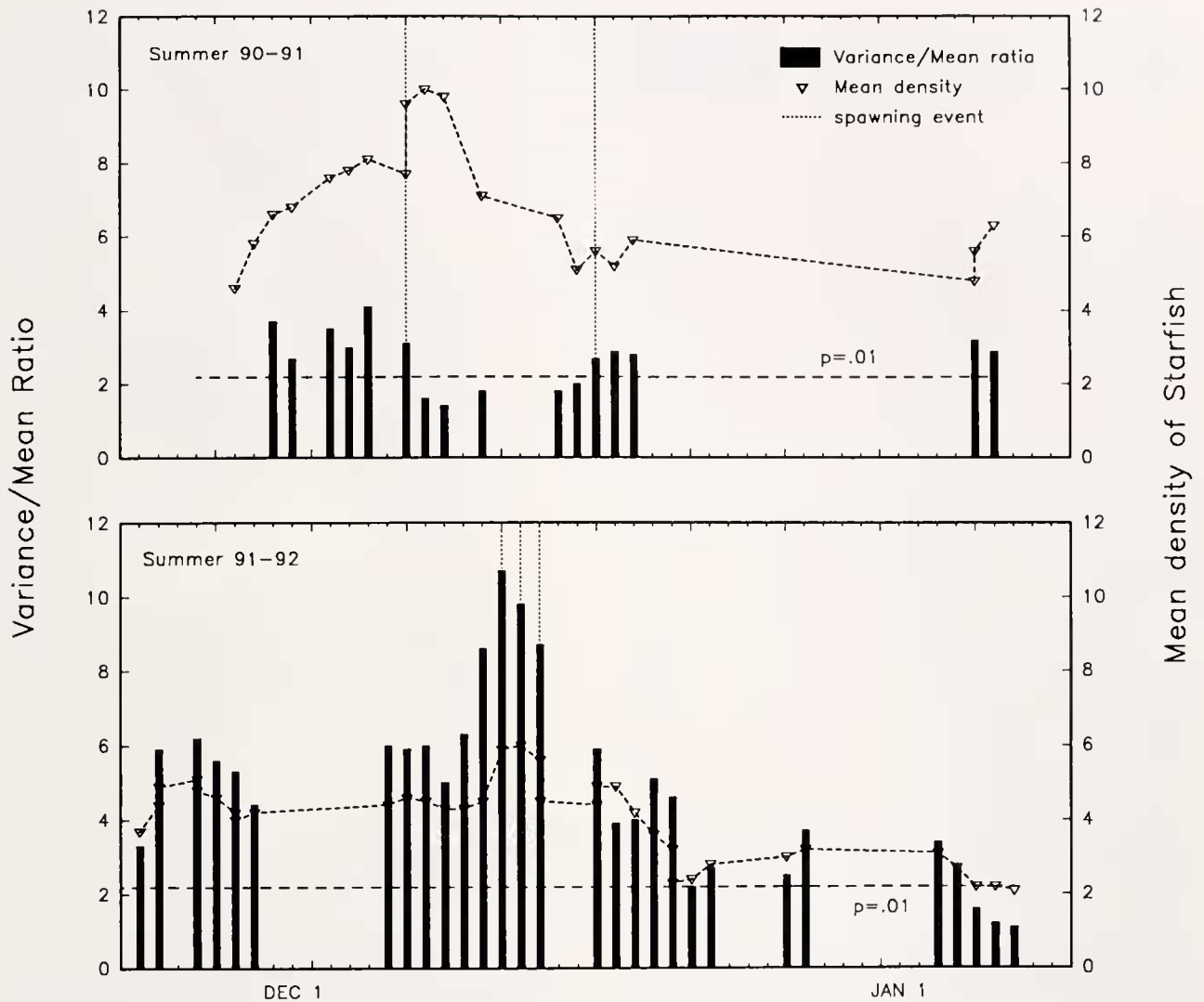


Figure 1. Starfish number and degree of aggregation for *Acanthaster planci* at Davies Reef. Variance/mean ratios (aggregation index) are calculated from counts of starfish along a 200×10 m belt transect during December and January of 1990-91 and 1991-92. Data for both counts and aggregation index are presented as three-point running means. Dates of spawning are indicated by vertical dotted lines. The 0.01 significance level for the aggregation index is taken from the table of Chi-square values; $df = 9$. Dates of observed natural spawnings: 1991; Dec 7 (38 females, 50 males), Dec 17 (three males). 1992; Dec 11 (1 female, 2 males), Dec 12 (2 females, 6 males), Dec 13 (1 male). 1992; Jan 23 (2 males [not included in figure]).

above the substratum to avoid any significant velocity gradient. The model assumes a smooth bottom and is unable to deal with the complex flow around corals that intrude into the flow. Such corals tend to slow and redirect the flow passing around them, generating turbulence. The turbulence would have the beneficial effect, in terms of the model, of smoothing the flow velocity variation within the water column. For the purposes of the model, it is assumed that the "bottom" is situated at a theoretical level just above the region where most of the coralline disturbance to the flow takes place. We also assume that

both the male and female are positioned 0.5 m above this bottom. In terms of the experiment, this assumption is reasonable because the spawning starfish were placed on top of outcroppings, at heights between 0.5 and 1.5 m above the substratum.

The field site for the fertilization experiments was a 100 m channel, about 7 m deep and 35 m wide, between two large patch reefs within the lagoon at Davies Reef. The bottom was sandy, with many coral outcroppings between 0.5 m and 2 m in height. The bottom of the site at Sesoko Island was similar; however the site was at the

Table 1

Field fertilization rates for *A. planci*. Percent fertilization, plus or minus standard error, and sample size (*n*) for experimentally induced spawnings in which eggs were collected at varying distances from spawning male starfish

Distance male to female (m)	Davies Reef						Sesoko Island
	Single male					5 males	Single male
	Distance offset from directly downstream (m)						
	0	2	4	8	16	0	0
-8	0.7 ^{±3.3} (6)	—	—	—	—	2.5 ^{±1.5} (8)	—
-4	16.1 ^{±3.0} (50)	—	—	—	—	—	—
0	90.1 ^{±2.1} (60)	—	0.5 ^{±0.9} (4)	0	0	98.7 ^{±0.4} (9)	75.6 ^{±1.9} (27)
2	86.5 ^{±2.6} (12)	—	—	—	—	—	47.2 ^{±10.3} (9)
4	71.8 ^{±8.4} (10)	10.7 ^{±5.7} (2)	0	0	—	—	48.6 ^{±5.0} (21)
8	71.9 ^{±9.5} (15)	80.3 ^{±7.6} (10)	29.5 ^{±8.1} (19)	—	—	—	38.9 ^{±4.3} (24)
16	41.5 ^{±5.2} (32)	—	39.1 ^{±11.7} (10)	4.1 ^{±2.1} (18)	—	33.7 ^{±4.7} (10)	25.9 ^{±3.8} (21)
32	26.8 ^{±3.9} (37)	—	31.9 ^{±10.3} (10)	13.8 ^{±7.6} (10)	2.6 ^{±1.4} (10)	30.2 ^{±5.0} (10)	16.7 ^{±3.9} (12)
64	20.5 ^{±5.8} (16)	—	—	—	—	23.8 ^{±5.0} (9)	—
100	5.8 ^{±1.4} (11)	—	—	—	—	19.6 ^{±4.8} (11)	—

Positive values indicate downstream positions; negative values indicate female starfish upstream of spawning male.

bottom of the outer reef slope (5 m), with currents traveling parallel to the reef front and thus bounded only on one side.

Results

Behavioral observations

We observed natural spawnings of *Acanthaster* on six occasions during the summers of 1990–91 and 1991–92 (Fig. 1). Of the four spawnings seen in 1991–92, females released eggs on only one occasion, 12 December 1991, when eight starfish (six male and two female) were observed, probably at the end of a spawning event involving many more animals. Fertilization rates in the eggs sampled directly downstream from a spawning female averaged 99%. We obtained egg samples from an additional two spawnings seen in 1990. Fertilizations ranged from 83%, during the peak of a large spawning, to 23%, at the end of spawning or during a minor spawning (details in Babcock and Mundy, 1992). Density of starfish on the study site and the degree of aggregation varied during each

spawning season (Fig. 1). In both seasons the highest densities of starfish were counted on and around times when major spawnings were seen. It was not possible to determine where the additional animals came from, but it is likely that the changes in abundance were a result of animals being less cryptic around the time of spawning, rather than any more specific movement of the population. Variance/mean ratios were higher in the weeks prior to or during the first major spawnings, indicating a greater tendency to aggregate, than they were later in the season when most of the spawning had already taken place. Although the population showed a significant level of aggregation during most of the surveys, the frequency of such observations was lower after spawning.

Induced spawning experiments

Fertilization rates for single female starfish at Davies Reef were still at detectable levels (5.8%) 100 m directly downstream from spawning male starfish, and were higher than 20% at 32 m and 64 m. For adjacent starfish, mean fertilization rates were 90.3%, but dropped to 16% at 4 m up-

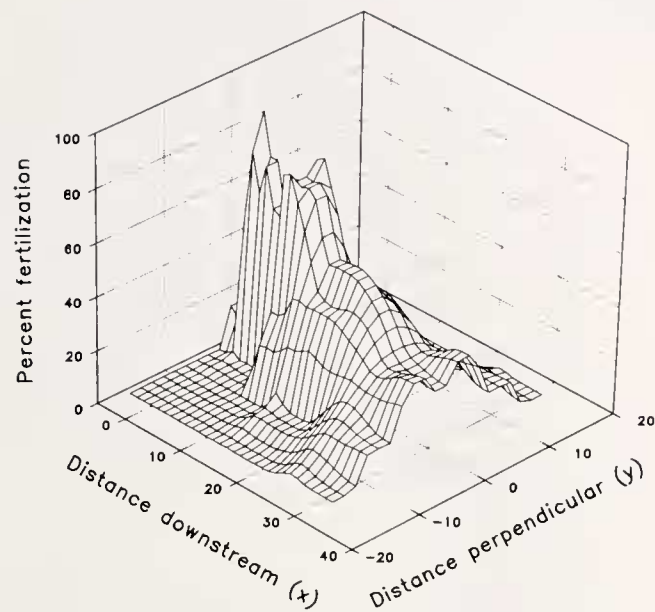


Figure 2. Three-dimensional representation of observed mean fertilization rates for *Acanthaster planci* at Davies Reef. Values used were means obtained from Table I for variations in fertilization at a range of coordinates surrounding a spawning male starfish. Current flows from left to right along the downstream (x) axis. (Missing data points interpolated using graphics features of SigmaPlot 5.)

stream, and to 0.7% at 8 m upstream (Table I). Fertilization rates dropped more rapidly as female starfish were moved away from the direct downstream (x) axis (Fig. 2). For example, no fertilization was recorded at 4 m downstream and 4 m normal, but at 8 m downstream, the sperm plume had diffused outward to the extent that mean fertilization was 29.5% at 4 m normal to the downstream axis. At 32 m downstream, fertilization was still measurable (2.6%) at 16 m perpendicular to the direct downstream axis, rising to 13.8% at 8 m and 31.9% at 4 m perpendicular.

For trials with more than one starfish, fertilization rates were expected to be higher than for single starfish at corresponding distances (Table I). This was not always the case, however, partly because these trials were conducted later in the season when the starfish had smaller gonads and partly because of the variability related to smaller sample sizes. Nevertheless, the average fertilization rate at 100 m downstream was 21.8% when five male starfish were induced to spawn. The maximum fertilization rate recorded at 100 m in trials with five males was 42%. Fertilization rates from starfish at Sesoko Island were lower than those from starfish at Davies Reef at equivalent distances downstream (Table I).

Effects of current speed on observed fertilization rates were slight and not significant over the range of current speeds experienced (Stepwise regression. Distance downstream: partial $r^2 = 0.521$, $F = 129.5$, $p = .0001$. Current

velocity: partial $r^2 = .014$, $F = 3.5$, $p = .063$). Trials in which currents reversed during the course of the experiment were not included in this analysis.

Sperm diffusion modeling

Diffusion coefficients. In order to determine the plume coefficients α_z , α_y and β , we concentrated on matching the model prediction to the results recorded along the axis 4 m ($y = 4$) to the side of the flow axis (Table I). By fitting the model to this more complex data set, which contained rising then falling values, we were able to obtain a more satisfactory set of coefficients to describe the diffusion of the plume than if we had used $y = 0$.

The model's predictions were found to be most sensitive to the β value. In general, the lower the β value the flatter the curve and the closer the sets of parameters matched. Because the range of values suggested by Denny and Shibata was for a model without a sea-surface boundary, we decided that a small deviation from these values would be acceptable. Values of $\beta = 0.5$, $\alpha_z = 0.65$, and $\alpha_y = 1.15$ were selected as the set of parameters that best matched the model to the data while remaining within an acceptable range for the values. Levels of turbulence and diffusion experienced in the lagoon of Davies Reef are likely to be lower than those used by Denny and Shibata (1989), whose model described extremely turbulent surf zones.

Separation distance (x , y). Comparing the model to the measured data (Fig. 3) reveals a fit that we believe is a reasonable enough representation of actual sperm diffusion to permit discussion of the model's behavior in more general terms. At short distances from the origin, predicted values were slightly higher than those we observed in our samples. This discrepancy could be an artifact of the use of induced spawnings if some of the eggs that were shed

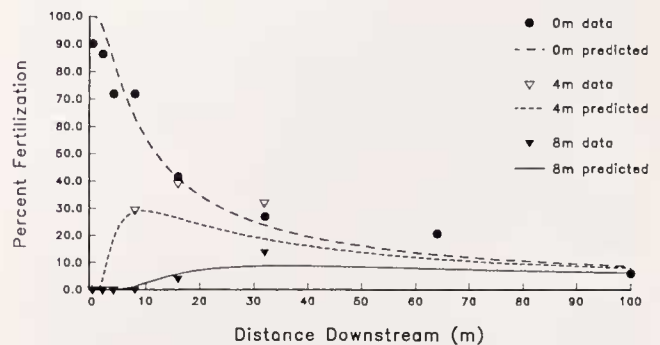


Figure 3. Observed and predicted mean fertilization rates for starfish at Davies Reef. Data points are from Table I, 0, 4, and 8 m offset from direct downstream axis. Predicted values are those derived using values that provided the best fit to data 4 m offset from x axis ($y = 4$). $\alpha_y = 1.15$; $\alpha_z = 0.65$; $\beta = 0.5$; $Q_s = 6.14 \times 10^8$; $\phi = 9.42 \times 10^{-10}$; $U = 0.12 \text{ m} \cdot \text{s}^{-1}$; $u^* = 0.1$; Depth = 7 m; release height $h = 0.5 \text{ m}$; vertical separation $z = 0.5 \text{ m}$.

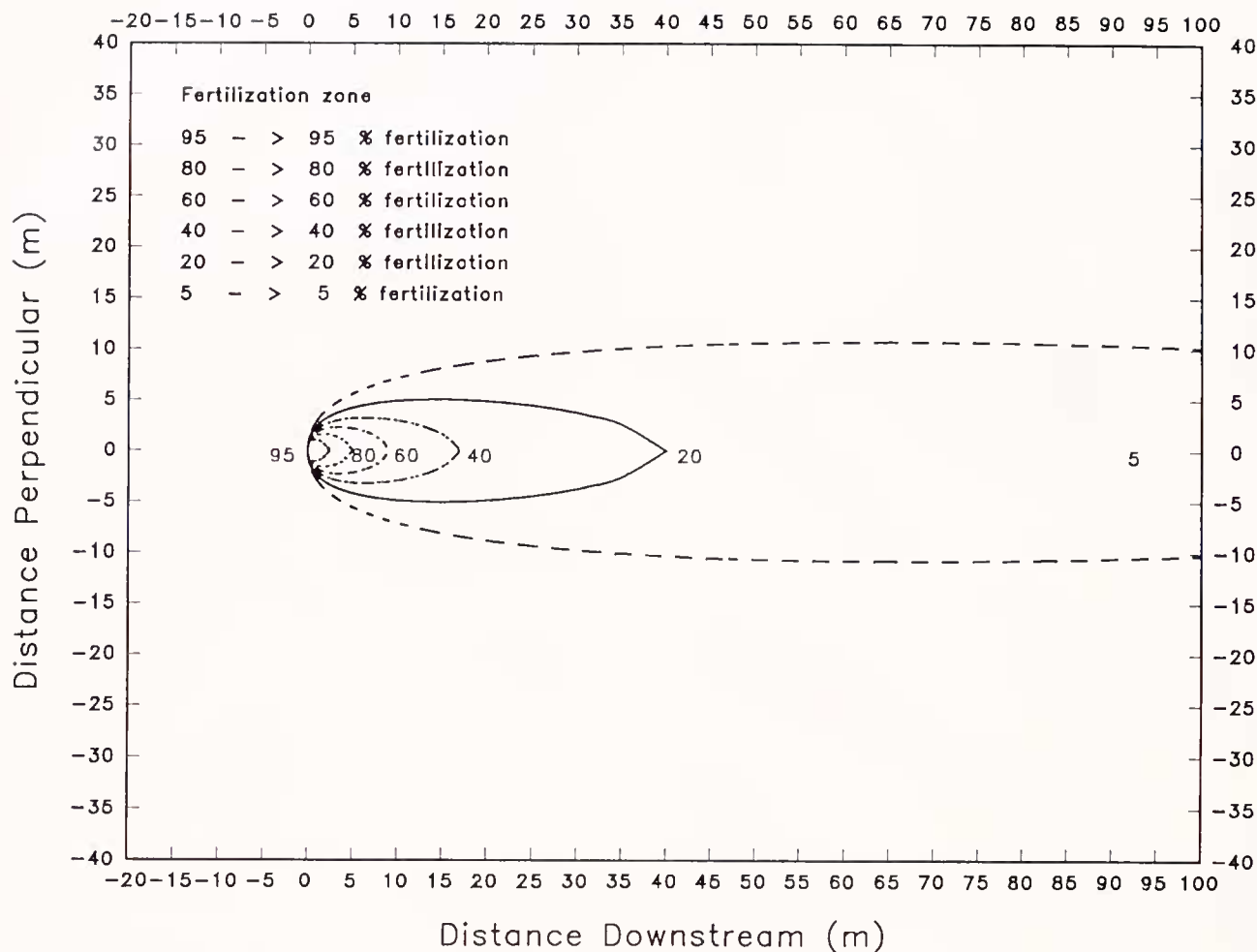


Figure 4. Fertilization contours for eggs released in a field downstream from a spawning male starfish. Parameters for model as for Figure 3. Y-coordinates for different fertilization rates were determined by substituting the relevant levels of fertilization in Eq. 5, and solving for y . Current flow is along the x axis from left to right. Fertilization zone contours correspond to $5 > 5\%$, $20 > 20\%$, $40 > 40\%$, $60 > 60\%$, $80 > 80\%$, and $95 > 95\%$.

were not mature and lacked the potential to be fertilized; inhibition of fertilization by high concentrations of fluorescein is also a possibility (Finkel *et al.*, 1981). Further from the origin the predicted values are generally lower than observed values. We also predicted fertilization success in two dimensions over the floor of the reef, producing contour maps of predicted fertilization success (Fig. 4). According to the model, fertilization will be greater than 5% within a long narrow plume region reaching out past 100 m downstream of the source, but the plume will only spread to a distance of about 15 m each side of the source. At 30 m downstream, 20–40% of the eggs of a spawning female would be fertilized, and at downstream separations less than 5 m, more than 80% of the ova would be fertilized, indicating that the success of spawning exhibits relatively low sensitivity to downstream separation when the starfish are aligned along the flow axis. These predictions

agree well with our observations (Table 1), providing an excellent correlation between observed means for field data and median values for predicted data points (Spearman rank correlation $r_s = .82$, $p < .0005$, $n = 23$).

Water depth (z). In water shallower than 20 m, where *A. planci* are likely to be found, the plume is confined between the surface and the sea floor. As the depth of water increases, the plume is able to disperse through a greater volume, and reflection of sperm from the surface takes longer to occur. Because of this, vertical separation of spawning animals will have an increasingly detrimental effect on fertilization success as depth increases (Fig. 5). In an animal such as *Acanthaster* that has the capacity to effect fertilization at distances that are large relative to total water depth, the presence of a term describing reflection from the surface is essential to an understanding of the effect of water depth on fertilization rate.

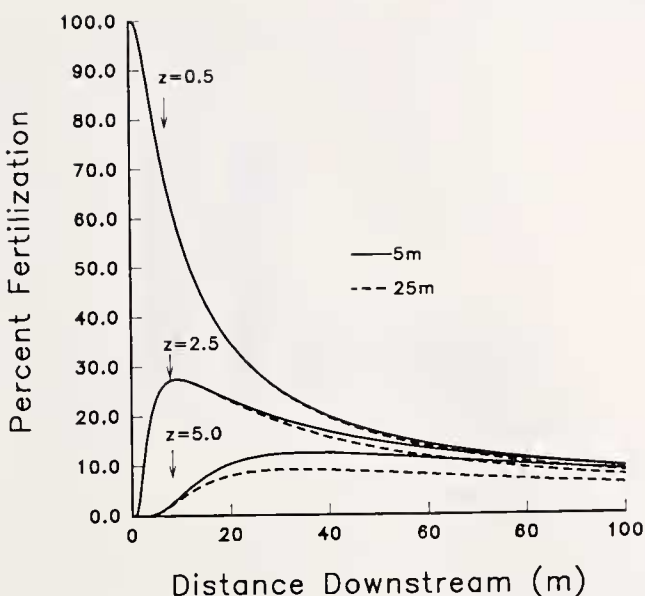


Figure 5. Predicted effects of depth and vertical separation on predicted fertilization rates. Diffusion parameters as for Figure 3.

Sperm release rate (Q_s). The phenomenal fertilization success of *A. planci* can be directly attributed to the animal's high sperm output. By examining the effect of differing sperm outputs, we examined how the spawning of more than one animal, or reduced levels of sperm production, could determine the success of external fertilization. The importance of sperm output level to fertilization success is clearly visible in the comparison of animals from Sesoko Island with those from Davies Reef. When sperm release values in the model were reduced so that they would correspond to those observed for starfish at Sesoko Island, there was a high level of agreement between the predictions and observed fertilization rates (Fig. 6). A group of five spawning male starfish is predicted, using the same model, to be capable of generating fertilization rates in excess of 40% at distances of 100 m or more downstream. Indeed the highest mean fertilization rate we observed for a group of five spawning males was 42% at 100 m (although the mean was much lower, 19.6%). If we compare these observations to the predictions of the model based on the gamete release rate of urchins such as *Strongylocentrotus* (1–3 g, Denny and Shibata, 1989), it becomes apparent why *A. planci* are capable of fertilization at distances much greater than those that are possible for urchins. Sperm release rate can clearly be a major factor determining the outcome of reproductive behavior in a free-spawning marine organism such as *Acanthaster*.

Discussion

Free-spawning marine invertebrates can and do achieve high levels of fertilization during normal spawning events,

as confirmed by results presented here as well as by those of other studies (Babcock and Mundy, 1992). In some spawnings of *A. planci*, success can be low, but the majority of gametes are shed during events in which the probability of fertilization is high (Babcock and Mundy, 1992). Levels of synchrony in the spawnings we observed were variable, and most spawnings involved only male starfish; such behavior may be wasteful of sperm. The release of eggs appeared to be more critically controlled, occurring only when many males were spawning. On the only such occasion observed in 1991–92, more than 99% fertilization was recorded for eggs released at the peak of spawning. Similar measurements made by Babcock and Mundy (1992) during 1990–91 indicate that spawning at the peak of a reproductive event involving a large proportion of the population will produce measurably higher fertilization rates (83%) than spawning at the end of the same event, or on occasions when only a small proportion of the population is spawning (~23%). Variations in spawning synchrony have been shown to produce corresponding changes in fertilization rate for holothurians (Babcock *et al.*, 1992) as well as for some mass-spawning corals (Oliver and Babcock, 1992).

Although members of the population may spawn several times, the relative contribution of each spawning event to the seasonal reproductive output probably varies considerably. Not only is the number of gametes released dependent on the numbers of animals participating and the intensity of spawning, but the probability of fertilization also varies in direct proportion to these factors. Thus variations in sperm release rate that would result from changes in synchrony are analogous to the critical impact of sperm release rate on sperm concentration and ultimately on fertilization (Fig. 6). In *Acanthaster*, for example, even though the reproductive season may be as long as two months and animals may spawn several times, the bulk of larvae may be produced in just one large reproductive event. The likelihood that particular events contribute disproportionately to reproductive output is increased by the presence of seasonal changes in the fertility and viability of gametes during the course of the season (Babcock and Mundy, 1994).

Adult aggregative behaviors in free-spawning invertebrates can also act to increase sperm concentrations at the time of spawning (Leviton, 1991). It is more likely that males in aggregations will shed sperm close to females where the sperm will have a better chance of fertilizing eggs. Aggregative spawning behavior has been clearly observed in other asteroids (*e.g.*, Minchin, 1987), but if *Acanthaster* has some means of aggregating at the time of spawning (Beach *et al.*, 1975), the intensity of this aggregation is not great (Fig. 1). Increases in the density of the population at Davies Reef indicated a change of starfish behavior and may represent an increase in aggregation

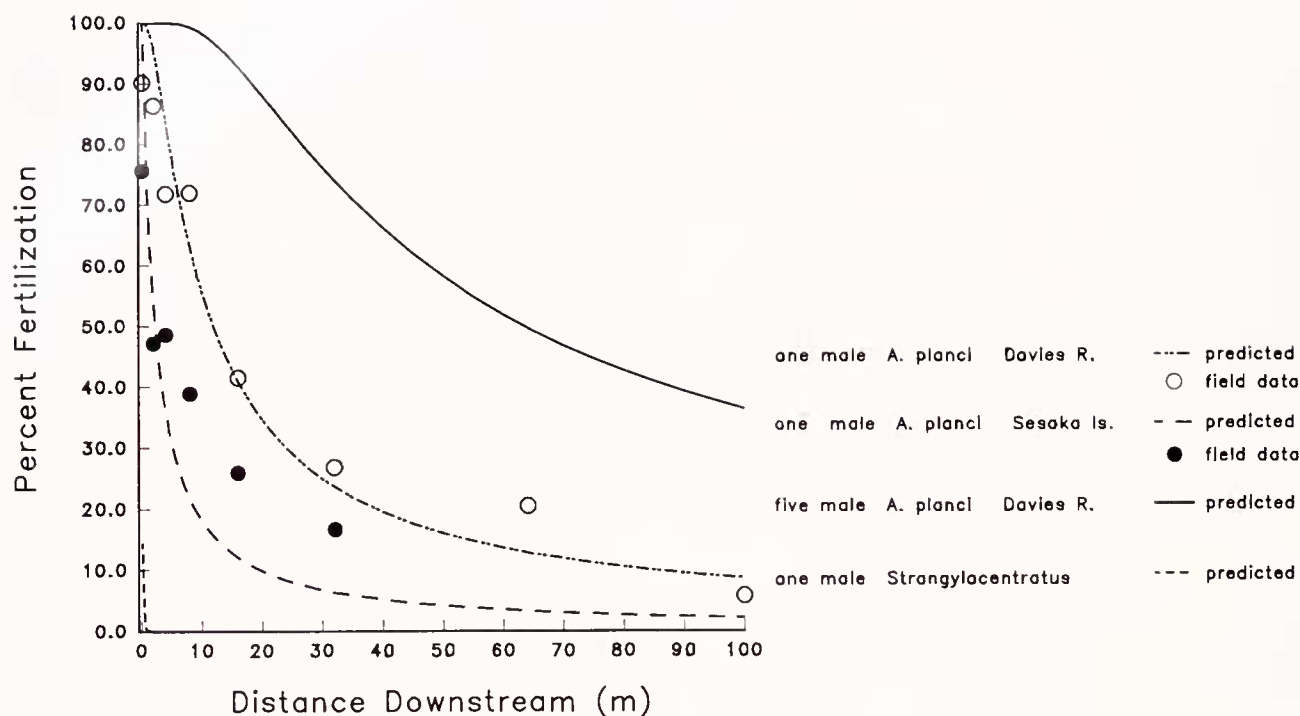


Figure 6. Effects of varying sperm release rate on predicted fertilization rate. Predictions and data for values on the direct downstream axis ($y = 0$). Model parameters for *Acanthaster planci* as for Figure 3. Sperm release rates: 1 male, Davies R. = 6.14×10^8 ; 5 males, Davies R. = 3.07×10^9 ; 1 male, Sesoko Is. = 1.5×10^8 ; 1 *Strongylocentrotus* = 1×10^7 ; all other parameters for estimates of *Strongylocentrotus* fertilization as in Denny and Shibata (1989), except $\alpha_y = 1.24$ and $\alpha_z = 1.98$.

on a scale too large for our sampling to detect. The fact that *Acanthaster* routinely aggregates in response to both the physical structure of the habitat (*e.g.*, cryptic resting places [personal obs.]) and the feeding of other starfish (Ormond *et al.*, 1973) does not preclude the usefulness of such behavior in terms of reproductive success.

The number of zygotes formed at relatively large distances from a sperm source may be high, as has been noted by Grosberg (1991), due to an increase in the total area under consideration, even though the proportion of eggs fertilized is lower than that in the small area immediately surrounding the source. This is an important factor in reproductive success not only at the population level but also in terms of the pattern of gene flow of individual organisms. For individual starfish, a high level of aggregation may not be necessary to optimize the transfer of genes to the next generation; despite the high level of fertilization for animals in close proximity, the total numbers of eggs fertilized by the sperm of a single spawning male may actually be greater at considerable distances from the site of sperm release (Fig. 7). As long as other members of the population are spawning in the vicinity and the release of gametes is essentially simultaneous or epidemic, intense aggregation is not essential to ensure significant fertilization success for this animal. It is interesting to

note that outbreaking populations of starfish may be better able to synchronise their spawning behavior (Okaji, 1991), probably through spawning pheromones such as those demonstrated by Miller (1989).

Other behaviors associated with spawning may act to increase the probability of spawning. *Acanthaster* populations have been observed to move into shallow water at the time of spawning (*e.g.*, Owens, 1971), as have populations of other asteroids (Minchin, 1987). In animals that live along coastlines or other steeply sloping substrata, this movement will have the effect of aggregating the population, reducing the distance separating the population in the horizontal as well as the vertical dimension. In addition, the dilution of gametes will be reduced in shallow water, due to reflection and constraint by the surface (Fig. 5). An extreme example of this phenomenon may be the reproductive behavior of polychaetes whose epitokes aggregate at the surface (*e.g.*, Palolo worms; Caspers, 1984), or hermaphroditic corals that shed buoyant gamete bundles that break apart at the surface (Oliver and Babcock, 1992).

Large-scale movements of populations may be true migrations of a sort, but they may also be the consequence of simple climbing behaviors. On a smaller scale, starfish commonly arch themselves up off the bottom when



Figure 7. Number of zygotes formed at varying distances from sperm source. Values are those predicted for the case of a single male *Acanthaster planci* spawning in a field of uniformly distributed female starfish. Density of female starfish is taken to be half the mean density of starfish on our study transect at Davies Reef during 1990–92, or 0.0124 female starfish $\cdot$ m 2 . Based on our gonad index data (Babcock and Mundy, 1992; and unpublished data) each of these females could spawn an average of 220 g of eggs during a natural spawning event. At approximately 9×10^4 eggs $\cdot$ g $^{-1}$ (Conand, 1985), this gives an egg release of 1.98×10^7 per female or 2.46×10^5 eggs $\cdot$ m 2 . The area defined by each fertilization zone (cf. Fig. 4) was then multiplied by this value to provide a total egg production value in each zone. The number of zygotes produced in each zone was calculated as the product of the total egg production and the median value for fertilization rate in each zone. Zygote production was not calculated for the area in which fertilization rates were predicted to be less than 5%.

spawning, and similar behaviors (*i.e.*, rearing) are seen in holothurians (McEuen, 1988). We frequently observed such behaviors in spawning *Acanthaster*, as have many other authors (Pearson and Endean, 1969). This activity is commonly associated with spawning in various benthic marine animals (McEuen, 1988). In some situations, gametes can actually pile up on the substratum adjacent to spawning animals (Minchin, 1987). Such situations are unlikely to result in high levels of fertilization, and their consequences would be reduced by arching behavior that would raise the gonopores further into the flow field.

The results of field experiments on the fertilization success of *A. planci* spawnings indicate a high level of success at separations as great as 100 m. At first glance these results appear to contradict the results of empirical field studies (*e.g.*, Pennington, 1985), as well as existing models of sperm diffusion for sea urchins (Denny and Shibata, 1989), which predicted low success even at small separations. Here we examine the model under spawning conditions similar to those of *A. planci*. Given the fertilization success of *A. planci* recorded during field experiments, the question is whether turbulent diffusion alone can bring sperm and ova together over large source separations. Given the large volumes of sperm released by *Acanthaster*,

we conclude that turbulent diffusion is a sufficient mechanism to enable this animal to achieve the observed levels of fertilization success. The level of fertilization recorded in our trials generally varied according to the mass of gonads available to be spawned. Fertilization was thus appreciably lower for the smaller Sesoko Island starfish than for Davies Reef starfish, and the levels observed were consistent with the predictions of our sperm diffusion model. This result contrasts with the conclusions of Levitan (1991) that body size and the amount of gametes released were not significant factors in fertilization success. This difference may have several sources. Firstly, the numbers of sperm released may not have been sufficiently large to produce a consistent difference with size, despite the relative difference in gonad volume between large and small urchins. Secondly, low current speeds and the short lifespan of urchin sperm relative to that of *Acanthaster* (J. Benzie, unpublished data) may also have contributed. We demonstrated that the same model that has been used to describe the rapid dilution of gametes and decline in fertilization success in echinoids is also consistent with our results, and that it is the exceptionally high rate of gamete release that, so far, sets this animal apart. Smaller organisms may be more reliant on phenomena such as aggregation, or even pseudo-copulation (*e.g.*, *Archaster typicus*; Run *et al.*, 1988), to ensure fertilization.

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